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CLAIMS

1. A method for determination of an analyte in a sample in a flow matrix by use of a transport flow of one or more biospecific affinity reactants, at least one of which is analytically detectable (Reactant*) and one is firmly anchored in the matrix (Reactant I), and the flow matrix comprises:

A) an application zone for liquid (LZ), containing buffer and sample and optionally one or more of the reactants, but not Reactant I,

B) a detection zone (DZ) located downstream of LZ with the firmly anchored reactant (Reactant I), and

C) optionally one or more zones in which any of the reactants has been pre-deposited,

wherein (i) the flow towards the detection zone is initiated by addition of the liquid with sample in the application zone LZS for transport of analyte and reactants towards the detection zone (DZ), and (ii) the amount of Reactant* bound to DZ is detected, the detected amount being related to the amount of analyte in the sample,

a ~~characterized in that~~ wherein

I. the flow matrix comprises at least two application zones for liquid arranged substantially adjacent to each other:

LZ_m . . . LZ_n . . . LZ_1 DZ

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wherein

a) LZ_n is an application zone for liquid, and n is the position of the application zone LZ_n,

b) m is the total number of application zones in which flow is initiated ($m \geq 2$),

c) one LZ_n is an application zone for sample (LZ_n,S) and one $LZ_{n''}$ is for Reactant* ($LZ_{n''}$.R*) with $n'' \geq n'$,

d) ---------------> is the direction of the flow, and

e) DZ is the detection zone, and

II. flow is initiated by adding liquid to each zone LZ_m . . LZ_n . . LZ_1 in such a way that $liquid_{n+1}$, added to the application zone LZ_{n+1}, is transported through the matrix immediately after $liquid_n$, added to the nearest downstream application zone LZ_n.

2. The method according to claim 1, ~~characterized in that~~ wherein $n'' > n'$ (sequential variants regarding analyte and Reactant*).

3. The method according to claim 1, ~~characterized in that~~ wherein $n'' = n'$ (simultaneous variants regarding analyte and Reactant*).

4. The method according to any of the claims 1 - 3, **characterized** in that Reactant* is pre-deposited in its application zone ($LZ_{n''}$.R*).

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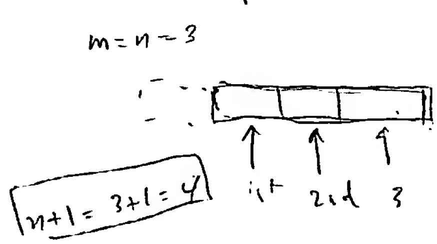
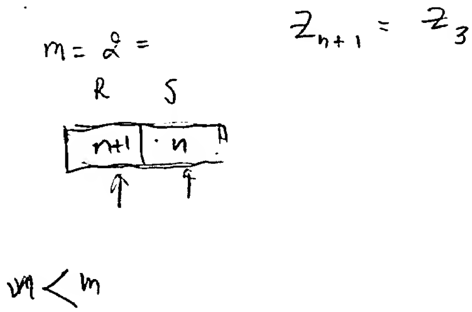
5. The method according to any of the claims 1 - 4, **characterized** in that $liquid_{n+1}$ is added to LZ_{n+1} before or substantially simultaneously with adding $liquid_n$ to LZ_n, with the exception of n = m, which zone lacks the zone LZ_{n+1}.





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6. The method according to any of the claims 1 - 5, **characterized** in that LZ_{n+1} finishes where LZ_n starts, with the exception of n = m, which zone lacks the zone LZ_{n+1}.

7. The method according to any of the claims 1 - 6, **characterized** in that application of liquid is performed substantially simultaneously in all LZ_m . . LZ_n . . LZ_1.

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8. The method according to any of the claims 1 - 7, **characterized** in that $m \leq 6$; n' is 1, 2 or 3; n'' > n'; $LZ_{n'+1}$, $LZ_{n'+2}$, $LZ_{n'+3}$, $LZ_{n''-1}$, and $LZ_{n''-2}$ are application zones for liquids intended for transport of Reactant* or other reactant or buffer without reactant, as far as allowed by m, n'' and n'.

9. The method according to any of the claims 1 - 8, **characterized** in that at least one of the zones LZ_m . . LZ_n . . LZ_1 comprises a pad or material layer applied on the flow matrix.

10. The method according to any of the claims 1 - 8, **characterized** in that the zones LZ_m . LZ_n . LZ_1 have zone spacers between each other.

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11. The method according to any of the claims 1 - 10, **characterized** in that the composition of transported components from an application zone LZ_n is not the same as from the nearest adjacent application zone LZ, in which flow is initiated, (LZ_{n+1} and LZ_{n-1}, with the exception of n = m and n = 1, which zones lack LZ_{n+1} and LZ_{n-1}, respectively).

12. The method according to any of the claims 1 - 11, **characterized** in that at least one reactant, other than Reactant*, is pre-deposited in an application zone LZ_n...R for liquid intended for transport of the reactant.

13. The method according to any of the claims 1 - 12, **characterized** in that m ≤ 6 and that n' for the application zone for sample ($LZ_{n'}S$) is 1, 2 or 3.

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14. The method according to any of the claims 1 - 13, **characterized** in that Reactant* has biospecific affinity for the analyte so that Reactant* is incorporated into a complex Reactant'---Analyte---Reactant* in the detection zone in an amount related to the amount of analyte in the sample, in which complex Reactant' has biospecific affinity to the analyte and is

(a) Reactant I, or

(b) a reactant to which Reactant I exhibits biospecific affinity and which is transported from $LZ_{n'}S$ or from an application zone downstream of $LZ_{n'}S$.

15. The method according to any of the claims 1 - 14, **characterized** in that the matrix comprises at least one calibrator zone (CZ), in which calibrator is bound to, or in advance has been bound to the matrix.

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16. The method according to claim ~~15~~, wherein ~~characterized in that~~ the calibrator zone or zones (CZ) have a binder for the calibrator firmly anchored in the matrix, the calibrator optionally being pre-deposited in the matrix upstream of the calibrator zone or zones.

17. The method according to any of the claims 1 - 16, **characterized** in that

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a. the analyte is chosen among antigens generally, and
b. the method is performed as part of diagnosing allergy or autoimmune disease.

18. A device for determination of an analyte in a sample in a flow matrix by use of a transport flow of one or more biospecific affinity reactants, at least one of which is analytically detectable (Reactant*) and one is firmly anchored in the matrix (Reactant I), said device comprising a flow matrix having:

A) an application zone for liquid (LZ), containing buffer and sample and optionally one or more of the reactants, but not Reactant I,

B) a detection zone (DZ) located downstream of LZ with the firmly anchored reactant (Reactant I), and

C) optionally one or more zones in which any of the reactants has been pre-deposited,

wherein
~~characterized in that~~

the flow matrix comprises at least two application zones for liquid arranged substantially adjacent to each other:

LZ_m . . . LZ_n . . . LZ_1 DZ

-->

wherein

a) LZ_n is an application zone for liquid, and n is the position of the application zone LZ_n,

b) m is the total number of application zones in which flow is initiated ($m \geq 2$),

c) one LZ_n is an application zone for sample (LZ_n,S) and one LZ_n is for Reactant* ($LZ_{n''}$.R*) with $n'' \geq n'$,

d) --------------> is the direction of the flow, and

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~~e) DZ is the detection zone.~~

19. The device according to claim 18, **characterized** in that n'' > n' and that the device is intended for sequential transport of analyte and Reactant*.

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20. The device according to claim 18, **characterized** in that n'' = n' and that the device is intended for simultaneous transport of analyte and Reactant*.

21 The device according to any of the claims 18 - 20, **characterized** in that Reactant* is pre-deposited in its application zone (LZ_n..R*).

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22. The device according to any of the claims 18 - 21, **characterized** in that LZ_{n+1} finishes where LZ_n starts, with the exception of n = m, which zone lacks the zone LZ_{n+1}.

23. The device according to any of the claims 18 - 22, **characterized** in that m ≤ 6; n' is 1, 2 or 3; n'' > n'; $LZ_{n'+1}$, $LZ_{n'+2}$, $LZ_{n'+3}$, $LZ_{n'-1}$, and $LZ_{n'-2}$ are application zones for liquids intended for transport of Reactant* or other reactant or buffer without reactant, as far as allowed by m, n'' and n'.

24. The device according to any of the claims 18 - 23, **characterized** in that the zones LZ_m . . LZ_n . . LZ_1 have zone spacers between each other.

25. The device according to any of the claims 18 - 23, **characterized** in that at least one of the zones LZ_m . . LZ_n . . LZ_1 comprises a pad or material layer applied on the flow matrix.

26. The device according to any of the claims 18 - 25, **characterized** in that at least one reactant, other than Reactant*, is pre-deposited in an application zone $LZ_{n'}$..R for liquid intended for transport of the reactant.

27. The device according to any of the claims 18 - 26, **characterized** in that m ≤ 6 and that n' for the application zone for sample ($LZ_{n'}$,S) is 1, 2 or 3.

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28. The device according to any of the claims 18 - 27, **characterized** in that the detection zone DZ comprises firmly anchored Reactant I, and that a reactant to which Reactant I exhibits biospecific affinity optionally is pre-deposited in LZ_n,S or in an application zone downstream of LZ_n,S.

29. The device according to any of the claims 18 - 28, **characterized** in that the flow matrix comprises at least one calibrator zone CZ, in which a calibrator or a binder for the calibrator is firmly anchored in the matrix.

30. The device according to claim 29, **characterized** in that the calibrator zone or zones (CZ) have a binder for the calibrator firmly anchored in the matrix, and that calibrator optionally is pre-deposited in the matrix upstream of the calibrator zone or zones.

31. The device according to any of the claims 18 - 30, **characterized** in that the device is intended for diagnosing allergy or autoimmune disease.

32. A test kit, **characterized** in that the kit comprises (i) a device according to any of claims 18 - 29 and (ii) Reactant*.

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33. The test kit according to claim 32, ~~characterized in that~~ wherein the kit additionally comprises (iii) a calibrator when a binder for the calibrator is firmly anchored in the matrix.